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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
09/044,696	03/18/98	BARCHFELD G	1393.002

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EXAMINER	
DEVI,S	
ART UNIT	**PAPER NUMBER**
1641	10

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Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary	Application No. 09/044,696	Applicant(s) Barchfeld et al.
	Examiner S. Devi, Ph.D.	Group Art Unit 1641

☒ Responsive to communication(s) filed on *Jul 6, 1999* .

☐ This action is **FINAL.**

☐ Since this application is in condition for allowance except for formal matters, **prosecution as to the merits is closed** in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

A shortened statutory period for response to this action is set to expire *three* month(s), or thirty days, whichever is longer, from the mailing date of this communication. Failure to respond within the period for response will cause the application to become abandoned. (35 U.S.C. § 133). Extensions of time may be obtained under the provisions of 37 CFR 1.136(a).

Disposition of Claims

☒ Claim(s) *1-30* is/~~are~~ pending in the application.

Of the above, claim(s) *1-18* is/~~are~~ withdrawn from consideration.

☐ Claim(s) _____ is/are allowed.

☒ Claim(s) *19-30* is/~~are~~ rejected.

☐ Claim(s) _____ is/are objected to.

☒ Claims *1-30* are subject to restriction or election requirement.

Application Papers

☒ See the attached Notice of Draftsperson's Patent Drawing Review, PTO-948.

☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

☐ The proposed drawing correction, filed on _____ is ☐approved ☐disapproved.

☐ The specification is objected to by the Examiner.

☒ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. § 119

☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d).

☐ All ☐ Some* ☐ None of the CERTIFIED copies of the priority documents have been

☐ received.

☐ received in Application No. (Series Code/Serial Number) _____ .

☐ received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

*Certified copies not received: _____ .

☒ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

Attachment(s)

☒ Notice of References Cited, PTO-892

☒ Information Disclosure Statement(s), PTO-1449, Paper No(s). *4 & 5*

☐ Interview Summary, PTO-413

☒ Notice of Draftsperson's Patent Drawing Review, PTO-948

☐ Notice of Informal Patent Application, PTO-152

— SEE OFFICE ACTION ON THE FOLLOWING PAGES —

DETAILED ACTION

Preliminary Amendments

1) Acknowledgment is made of Applicants' amendments filed 07/10/98 (paper no. 3) and 07/30/98 (paper no. 7). With these, Applicants have amended the specification and complied with the Sequence Rules.

Election

2) Acknowledgment is made of Applicants' election filed 07/06/99 (paper no. 9), without traverse, of invention III, claims 19-30, and of species, *E. coli* heat labile toxins LT-K63 and LT-R72, in response to the restriction requirement mailed 06/07/99 (paper no. 8). Upon further consideration, the requirement for species election made for the toxin species of claim 20 and the mutant species of claim 22, are withdrawn, and all species in these claims have been examined.

Claims Status

3) Claims 1-30 are pending in this application.

Claims 1-18 are withdrawn from further consideration by the Examiner of record, 37 C.F.R 1.142(b), as being drawn to a non-elected invention.

Elected claims 19-30 are under examination and an Action on the Merits for these claims is issued in the instant Office Action (paper no. 10).

Sequence Listing

4) Acknowledgment is made of Applicants' submission of Sequence Listing filed 07/30/98 (paper no. 7) which has been entered.

Information Disclosure Statements

5) Acknowledgment is made of Applicants' Information Disclosure Statements filed 05/14/98 (paper no. 4) and 09/14/98 (paper no. 5). The information referred to therein has been considered and a signed copy is attached to this Office Action (paper no. 10).

Domestic Priority

6) The instant application claims priority to a provisional application, SN 60/041,227, filed 03/21/97.

Drawings

7) The drawings are objected to under 37 C.F.R 1.84 because of the reasons set forth by the Draftsperson in the attached Form PTO 948. Correction is required.

Oath/Declaration

8) The oath or declaration is defective. A new oath or declaration in compliance with 37 C.F.R 1.67(a) identifying this application by application number and filing date is required. See MEP. §§ 602.01 and 602.02.

The oath or declaration is defective because it is not executed in accordance with either 37 C.F.R 1.66 or 1.68.

Claims Rejections - 35 U.S.C. § 112, First Paragraph

9) Claim 22 is rejected under 35 U.S.C. 112, first paragraph, because the specification while being enabling for a method for immunizing a vertebrate subject comprising parenterally administering an effective amount of an adjuvant comprising a detoxified mutant of a bacterial ADP-ribosylating toxin, LT-K63 or PT-K9/G129, and at least one selected antigen, does not reasonably provide enablement for such a method using the adjuvant **detoxified** mutant species, CT-S109 or LT-R72 , as instantly claimed. It is apparent that the various "detoxified" mutant species of toxins recited in the instant claim are required to practice the full scope of the invention. The specification enables the construction and evaluation of LT-K63 and LT-R72 mutants (see working Examples 1-5 and Table 6) as parenteral adjuvants. The construction and evaluation of PT-K9/G129 as an adjuvant is known in the art and therefore a method of immunization using this mutant as an adjuvant is enabled. However, the two mutant species, CT-S109 or LT-R72, recited in the claim do not meet the enablement provision of 35 U.S.C. § 112, first paragraph, for the following reasons.

Many of the factors regarding undue experimentation have been summarized in *In re Wands*, 858 F.2d 731, 8 USPQ2d 1400 (Fed. Circ. 1988) as follows:

- The quantity of experimentation necessary (time and expense);
- The amount of direction or guidance presented;
- The presence or absence of working examples of the invention;
- The nature of the invention;
- The state of the art;

  The relative skill of those in the art;

- The predictability or unpredictability of the art; and
- The breadth of the claims.

In the instant case, although the construction and evaluation as an adjuvant of LT-R72 is described in the specification, the state of the art suggests that an LT mutant having a site directed substitution of Alanine to Arginine at position 72 is not "detoxified" as claimed currently. For instance, Pizza *et al.* (*Mol. Microbiol.* 14: 51-60, October 1994) teach such a mutant. See Table 1, Fig 1k and page 57, left column, last paragraph. Table 1 illustrates that the mutant named "R72" having a Ala->Arg sub A mutation remains toxic (toxicity is rated as ++). Pizza *et al.* state that this mutant was "fully toxic" as in the case of the mutant having a substitution at Ser-68 (see page 57, left column, last paragraph). Clearly, LT-R72 is not detoxified as claimed in instant claim 22.

With regard to the mutant CT-S109 recited in the instant claim, while there is a descriptive support in the instant application for a CT mutant comprising a substitution of serine at position 109 (CT-S109), there is no evidentiary support that such a mutant, if constructed, would be detoxified and functional as an adjuvant. It appears that there is considerable unpredictability in the art as to which of the site directed substitutions at specific positions would eliminate toxicity and at the same time retain structural and functional integrity of proteins. For instance, Pizza *et al.* (*Mol. Microbiol.* 14: 51-60, October 1994) teach that replacement of His-70, Val-60, Ala-45 and Leu-41 in *E. coli* LT-A resulted in the "collapse of the protein structures" and altered "the structural assembly" (see page 54, last two lines, and page 57) of the proteins. Furthermore, amino acid substitutions that resulted in mutant proteins, M59, H72 and N192, show that these mutants remained as toxic (+++) as the wild type toxin (see Table 1).

Undue experimentation would have been required by one of ordinary skill in the art at the time of the effective filing date of the instant application to reproducibly practice the invention as claimed due to the lack of specific guidance, the lack of working examples enabling detoxified and functional (as parenteral adjuvants) mutant species, LT-R72 and CT-S109 , the breadth of claims, the demonstrated unpredictability as reflected in the state of the art, and the quantity of experimentation necessary.

Claims Rejections - 35 U.S.C. § 112, Second Paragraph

10) Claims 21-24 are rejected under 35 U.S.C. §112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which Applicants regard as the invention.

(a) Claim 21 lacks antecedent basis for the recitation "the bacterial holotoxin". Claim 21 depends from claim 20, which recites a "toxin", but not "holotoxin".

(b) Claims 25 and 27 lack proper antecedent basis for the recitation "antigen". For clarity and proper antecedence, it is suggested that Applicants replace the recitation with --the antigen--.

(c) Claim 20 lacks antecedence for the recitation "the non-toxic". Claim 20 depends from 19 which recites "an adjuvant", but not "a non-toxic" adjuvant.

Claims Rejections - 35 U.S.C. § 102

11) The following is a quotation of the appropriate paragraphs of 35 U.S.C. § 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless --
>
> (a) the invention was known or used by others in this country, or patented or described in a printed publication in this or a foreign country, before the invention thereof by the applicant for a patent.
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

12) Claims 19-23 and 30 are rejected under 35 U.S.C § 102(b) as being anticipated by Tommaso *et al.* (*Infect. Immun.* 64: 974-979, 27 February 1996).

Tommaso *et al.* teach a method for immunizing mice by administering, by intravaginal route, microgram quantities of a selected antigen such as ova and an adjuvant comprising a detoxified mutant of an ADP-ribosylating heat-labile enterotoxin of *E. coli*, LTK63, (see abstract and page 975, left column). LTK63 is constructed by site-directed mutagenesis (see page 974) according to the teachings of Douce *et al.*, *PNAS* 92: 1644-1648, 1995, by replacing an amino acid at position 63 in the A subunit of the toxin (see pages 975 and 978).

Claims 19-23 and 30 are anticipated by Tommaso *et al.*

13) Claims 19-21, 25, 26 and 30 are rejected under 35 U.S.C § 102(b) as being anticipated by Douce *et al.* (*PNAS* 92:1644-1648, February 1995).

Douce *et al.* teach a method of immunizing mice (i.e. the vertebrate subject) by

administering microgram quantities (i.e. immunologically effective amount) of a selected antigen such as ova or fragment C of tetanus toxoid and non-toxic mutants of *E. coli* heat-labile toxin (LT) lacking ADP-ribosyltransferase activity and acting as non-toxic mucosal adjuvants (see title; abstract; Figures 2 and 3; Table 2, and page 1644, right column). The proteins are contained in phosphate-buffered saline (i.e. pharmaceutically acceptable or topical vehicle) (see page 1644). The detoxified mutant used for immunization comprises Arginine at position 7 of the A subunit replaced with Lysine by site-directed mutagenesis (see abstract). Mice are immunized by subcutaneous route with 10 micrograms of ova admixed with the non-toxic adjuvant LTK7, i.e. concurrent administration of the antigen and the adjuvant (see page 1645, right column).

Claims 19-21, 25, 26 and 30 are anticipated by Douce *et al.*

14) Claims 19-21 and 25-30 are rejected under 35 U.S.C § 102(b) as being anticipated by Rappuoli *et al.* (WO 95/17211, published 06/29/95 -Applicants' IDS) (WO '211).

Rappuoli *et al.* (WO '211) teach a method of immunizing mice (i.e. the vertebrate subject) by administering, subcutaneously, with microgram quantities (i.e. an immunologically effective amount) of an adjuvant comprising a detoxified mutant of a bacterial ADP-ribosylating toxin, i.e. LTK7 mutant derived from heat-labile toxin, LT, of *E. coli*, admixed with a selected antigen such as Fragment C of tetanus toxin or ovalbumin (see pages 14 and 16, last paragraphs). The detoxified mutant comprises one or more amino acid additions, deletions or substitutions in the A subunit of the holotoxin (see claim 4). Rappuoli *et al.* teach a method of immunization wherein the disclosed adjuvant and the antigen are administered parenterally, i.e., intramuscularly or transdermally, i.e. transcutaneously (see page 12, lines 8-13). The composition may contain "any" pharmaceutically acceptable carriers that do not themselves induce an immune response (see paragraph bridging page 9 and 10) and may contain water, saline, glycerol or ethanol (i.e. topical vehicles). Rappuoli *et al.* further teach a method of immunization wherein the adjuvant and the selected antigen are administered simultaneously (i.e. concurrently) or sequentially or separately, which encompass the methods recited in instant claims 30, 29 and 28 respectively (see claims 13 and 14 of Rappuoli *et al.*).

Claims 19-21 and 25-30 are anticipated by Rappuoli *et al.* (WO '211).

15) Claims 19-22, 25, 26 and 28-30 are rejected under 35 U.S.C § 102(b) as being anticipated

by Gajewzyk *et al.* (WO 95/34323, published 21 December 1995 - Applicants' IDS).

Gajewzyk *et al.* teach a method of immunizing mice or a human (i.e. the vertebrate subject) by administering a non-toxic adjuvant comprising a detoxified mutant of a *Bordetella pertussis* ADP-ribosylating toxin, K9G129 PT analogue and at least one selected antigen such as ovalbumin or a non-*Bordetella* antigen (see page 32, lines 14-16; page 11, lines 3-10, and claims 25, 28 and 29). The K9G129 PT analogue has two amino acids replaced (see page 10, lines 7-12). The two components are co-administered to the host in immunologically effective amounts (see claim 25). The term "coadministration" means simultaneous administration or administration within a few days (see page 11, lines 20-23). The composition may contain a physiologically acceptable carrier such as water, saline, glycerol or ethanol (see page 17, lines 8-11) and may be administered subcutaneously or intramuscularly (see page 17, lines 15-22). The genetically detoxified pertussis holotoxin has at least one amino acid removed (i.e. deletion) or replaced (i.e. substitution) in the holotoxin (see claim 12). The K9G129 adjuvant is evaluated with human pediatric combination vaccines, for example, a DTP vaccine (see page 15, lines 23-26 and page 13, lines 15-17).

Claims 19-22, 25, 26 and 28-30 are anticipated by Gajewzyk *et al.*

16) Claims 19-23 and 25-30 are rejected under 35 U.S.C § 102(a) as being anticipated by Pizza *et al.* (WO 97/02348, published 23 January 1997 -Applicants' IDS) (Pizza *et al.*, WO '348).

Pizza *et al.* (WO '348) disclose a method of immunizing a vertebrate subject by administering an immunologically effective amount or dose of a composition comprising a detoxified mutant protein of a bacterial toxin such as cholera toxin subunit A (CT-A) or *E. coli* heat-labile toxin subunit A (LT-A), (i.e. ADP-ribosylating toxins) comprising serine at position 63 and Arginine at position 192 replaced with another amino acid, a pharmaceutically acceptable carrier and a second immunogenic toxin antigen (see claims 11, 4, 2 and 1; page 15, lines 33-36, and page 16, lines 18 and 19). The pharmaceutically acceptable carriers or vehicles may be water, saline, glycerol or ethanol (see page 16, lines 1-5). The immunogenic detoxified protein acts as an adjuvant for the second immunogenic protein or antigen, administered separately, sequentially or along with the immunogenic detoxified protein (see page 9, lines 1, 2 and 9-13). One of the detoxified mutant protein used is LTK63 (see page 11). CTK63/G192, a detoxified mutant

protein derived from CT is also disclosed (see page 46). The composition may be conventionally administered parenterally, i.e. subcutaneously or intramuscularly, or may be administered transdermally, i.e. transcutaneously (see page 16, last paragraph).

Claims 19-23 and 25-30 are anticipated by Pizza *et al.* (WO '348).

Claims Rejections - 35 U.S.C. § 103

17) The following is a quotation of 35 U.S.C. § 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person. having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The factual inquiries set forth in *Graham v. John Deere Co.*, 148 USPQ 459, that are applied for establishing a background for determining obviousness under 35 U.S.C. § 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or unobviousness.

18) Claims 22-24 are rejected under 35 U.S.C. §103(a) as being unpatentable over Rappuoli *et al.* (WO 95/17211, published 06/29/95 -Applicants' IDS) (WO '211) as applied to claims 19-21 above, and further in view of Roberts *et al.* (*Infect. Immun.* 63: 2100-2108, June 1995), or Partidos *et al.* (*Immunology* 89: 483-487, December 1996).

The teachings of Rappuoli *et al.* (WO '211) have been explained above, which do not disclose the use of specific detoxified mutant species, LT-K63, LT-R72, CT-S109 and PT-K9/G129, recited as Markush species in the instant claims.

However, Roberts *et al.* teach a detoxified mutant of ADP-ribosylating pertussis toxin, PT-9K/129, as an adjuvant in a method of immunizing mice by administering the mutant in combination with phosphate-buffered saline and a selected antigen such as fragment C of tetanus toxin (see abstract and page 2101, left column). Roberts *et al.* also teach that PT-9K/129 itself is highly immunogenic in humans and has been licensed as a component of an acellular pertussis

vaccine in Italy (see page 2107).

Partidos *et al.* teach a detoxified mutant of ADP-ribosylating *E. coli* heat-labile toxin, LT-K63, as an adjuvant in a method of immunizing mice by administering the mutant in combination with phosphate-buffered saline and a selected antigen such as a measles viral antigen (see abstract; page 484, left column, and page 485, right column). The mutant adjuvant is an effective immunogen itself and produces LT-specific antibodies (see pages 483 and 486).

It would have been *prima facie* obvious to one skilled in the art at the time the invention was made to use Roberts' detoxified mutant of a ADP-ribosylating pertussis toxin, PT-9K/129, or Partidos' or Pizza's detoxified mutant of a ADP-ribosylating *E. coli* heat-labile toxin, LT-K63, as an alternative adjuvant having similar functions, in place of Rappuoli's LTK7 mutant in Rappuoli's method of immunizing mice by parenteral administration, to produce the instant invention for the expected benefit of also eliciting LT- or PT-specific protective antibodies as taught by Partidos *et al.* or Roberts *et al.*, in addition to enhancing the antibody response to the selected second antigen. A skilled artisan would have had a reasonable expectation of success in producing the instant invention since substituting one adjuvant comprising a detoxified mutant of an ADP-ribosylating toxin with an alternative, art-known, functionally equivalent detoxified mutant of ADP-ribosylating toxin would be expected to bring about similar, if not better, beneficial effects or results. Absent evidence to the contrary, the instant claims are obvious over the prior art of record.

Claims 22-24, as a whole, are obvious over the prior art of record.

Remarks

19) Claims 19-30 stand rejected.

20) The prior art made of record and not relied upon currently in any of the rejections is considered pertinent to Applicants' disclosure:

- Lobet *et al.* (*Infect. Immun.* 159: 2870-2879, 1991) teach mutant analogs of LT-A of *E. coli,* rLTA/R192G, produced by site-directed mutagenic alterations and their ADP-ribosyltransferase activity (see abstract).

Grant *et al.* (*Infect. Immun.* 62: 4270-4278, 1994) teach a recombinant LT-A produced by site-directed mutagenesis as a potential vaccine candidate.

Magagnoli *et al.* (*Infect. Immun.* 64: 5434-5438, December 1996) teach LT-K63 non-toxic mutant derivative of LT having a Ser-63->Lys mutation as a mucosal adjuvant and an immunogenic vaccine candidate (see abstract).

21) Papers related to this application may be submitted to Group 1600, AU 1641 by facsimile transmission. Papers should be transmitted via the PTO Fax Center located in Crystal Mall 1. The transmission of such papers by facsimile must conform with the notice published in the Official Gazette, 1096 OG 30, November 15, 1989. The CM1 facsimile center's telephone number is (703) 308-4242.

22) Any inquiry concerning this communication or earlier communications from the Examiner should be directed to S. Devi whose telephone number is (703) 308-9347. The Examiner can normally be reached on Monday to Friday from 8.00 am to 4.00 PM. A message may be left on the Examiner's voice mail system.

If attempts to reach the Examiner by telephone are unsuccessful, the Examiner's supervisor, James Housel, can be reached on (703) 308-4027. The fax phone number for this Group is (703) 305-7939.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the Group receptionist whose telephone number is (703) 308-0196.

August 1999

James C. Housel 8/2/99
JAMES C. HOUSEL
SUPERVISORY PATENT EXAMINER